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**Rotational thromboelastometry (ROTEM) parameters in dogs with haemoperitoneums and  
their association with clinical and laboratory signs**

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## **1. Zusammenfassung**

### **1.1 Summary**

Rotational thromboelastometry is a viscoelastic coagulation test that allows the evaluation of haemostasis from clot formation to clot dissolution. The aim of this retrospective study was to describe the changes in haemostasis using ROTEM parameters in dogs presenting with spontaneous or traumatic haemoperitoneum and to evaluate any associations between clinical and laboratory parameters at presentation with the ROTEM. We hypothesized that the dogs would show signs of hypocoagulability and hyperfibrinolysis and that these changes would correlate with the degree of hypoperfusion. Clinical records were searched for a period of 5 years for dogs presenting with a haemoperitoneum und for whom a ROTEM analysis at presentation was carried out. Forty dogs were identified, and various clinical and laboratory parameter were retrieved. The following ROTEM parameters were analysed: ExTEM CT, ExTEM CFT, ExTEM MCF and ExTEM ML, as well as FibTEM CT and MCF. Compared to institutional reference intervals, dogs with haemoperitoneum showed prolongation of ExTEM and FibTEM CT, ExTEM CFT and 50% were hypocoagulable and 62% thrombocytopenic. No hyperfibrinolysis could be detected. Statistics showed an association between decreased base excess, trauma and ROTEM signs for hypocoagulability. Furthermore, age was associated with a stronger fibrin clot. In conclusion, 50% of the dogs presented hypocoagulable and changes in ROTEM parameters are similar to those seen with consumption coagulopathy.

### **1.2 Zusammenfassung**

Als viskoelastisches Verfahren kann die Rotationsthromboelastometrie die Hämostase von der Gerinnselbildung bis hin zur -auflösung aufzeichnen. Das Ziel dieser retrospektiven Studie war es die Veränderungen in der Hämostase an Hand von ROTEM Parametern bei Hunden zu beschreiben, die mit spontanem oder traumatischem Hämoperitoneum vorgestellt wurden. Weiter

wurde der Zusammenhang zwischen den ROTEM Parametern und ausgewählten klinischen und Labor-Parametern untersucht. Wir stellten die Hypothese auf, dass die Hunde Zeichen von Hypokoagulabilität und Hyperfibrinolyse aufweisen, und diese Veränderungen mit der Ausprägung der Hypoperfusion korrelieren. Innerhalb des 5-jährigen Untersuchungsrahmens wurden vierzig Hunde mit Hämoperitoneum vorgestellt, für die bei Vorstellung eine ROTEM-Analyse durchgeführt wurde. Bei diesen Hunden wurden aus den Krankengeschichten klinische und Labor- Parameter herausgesucht. Die folgenden ROTEM Parameter wurden evaluiert: ExTEM CT, ExTEM CFT, ExTEM MCF und ExTEM ML sowie FibTEM CT und MCF. Die Hunde zeigten verlängerte ExTEM und FibTEM CT und verlängerte ExTEM CFT, 50% waren hypokoagulabel und 62% thrombozytopenisch. Hyperfibrinolyse konnte nicht dargestellt werden. Statistisch wurde ein Zusammenhang zwischen verminderten Basenüberschuss, Trauma und ROTEM Zeichen für Hypokoagulabilität dargestellt. Weiter war Alter mit einem stärkeren Fibrin-Gerinnsel assoziiert. Die aufgezeigten ROTEM Veränderungen entsprechen am ehesten einer Verbrauchskoagulopathie.

## **2. Abdruck des gedruckten Artikels**

# **Rotational thromboelastometry (ROTEM) parameters in dogs with haemoperitoneum and their associations with clinical and laboratory signs**

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# **Thromboelastometrie (ROTEM) Parameter und deren Assoziierung mit klinischen und Laborparametern in Hunden mit Hämoperitoneum**

Als viskoelastisches Verfahren kann die Rotationsthromboelastometrie (ROTEM) die Hämostase von der Gerinnungsbildung bis hin zur -auflösung aufzeichnen. Das Ziel dieser retrospektiven Studie war es die Veränderungen in der Hämostase an Hand von ROTEM Parametern bei Hunden zu beschreiben, die mit spontanem oder traumatischem Hämoperitoneum vorgestellt wurden. Weiter wurde der Zusammenhang zwischen den ROTEM Parametern und ausgewählten klinischen und Labor-Parametern untersucht. Wir stellten die Hypothese auf, dass die Hunde Zeichen von Hypokoagulabilität und Hyperfibrinolyse aufweisen, und diese Veränderungen mit der Ausprägung der Hypoperfusion korrelieren. Innerhalb des 5-jährigen Untersuchungsrahmens wurden vierzig Hunde mit Hämoperitoneum vorgestellt, für die bei Vorstellung eine ROTEM-Analyse durchgeführt wurde. Bei diesen Hunden wurden aus den Krankengeschichten klinische Parameter wie Herzfrequenz, Blutdruck, Glukose und Laktat, Serumalbumin, peripherer und abdominaler Hämatokrit, ionisiertes Kalzium, pH und Basenüberschuss herausgesucht. Die folgenden ROTEM Parameter wurden evaluiert: extrinsische Gerinnungszeit (ExTEM CT), Gerinnungsbildungszeit (ExTEM CFT), Gerinnungsstärke (ExTEM MCF) und maximale Lyse (ExTEM ML) sowie Fibrinogen (fib-tem) CT und MCF. Im Vergleich zu den Referenzintervallen zeigten die Hunde verlängerte ExTEM und FibTEM CT und verlängerte ExTEM CFT, 50% waren hypokoagulabel und 62% thrombozytopenisch. Hyperfibrinolyse konnte nicht dargestellt werden. Multiple lineare Regressionsmodelle haben einen Zusammenhang zwischen einem verminderten Basenüberschuss, Trauma und ROTEM Zeichen für Hypokoagulabilität dargestellt. Weiter war Alter mit einem stärkeren Fibrin-Gerinnung assoziiert.

Zusammenfassend präsentierten sich 50% der Hunde hypokoaguabel und die ROTEM Veränderungen entsprechen am ehesten einer Verbrauchskoagulopathie.

#### Schlüsselwörter

Canine, Hämoperitonium, Hämostase, Hyperfibrinolyse, Hypokoagulabilität



## Summary

Rotational thromboelastometry (ROTEM) is a viscoelastic coagulation test that allows the evaluation of haemostasis from clot formation to clot dissolution. The aim of this retrospective study was to describe the changes in haemostasis using ROTEM parameters in dogs presenting with spontaneous or traumatic haemoperitoneum and to evaluate any associations between clinical and laboratory parameters at presentation with the ROTEM. We hypothesized that the dogs would show signs of hypocoagulability and hyperfibrinolysis and that these changes would correlate with the degree of hypoperfusion. Clinical records were searched for a period of 5 years for dogs presenting with a haemoperitoneum and for whom a ROTEM analysis at presentation was carried out. Forty dogs were identified, and various clinical and laboratory parameters (heart rate, blood pressure, blood glucose, lactate, serum albumin concentration, PCV (venous and abdominal), ionized calcium, pH and base excess) were retrieved. The following ROTEM parameters were analysed: extrinsic clotting time (ExTEM CT), clot formation time (ExTEM CFT), clot firmness (ExTEM MCF) and maximum lysis (ExTEM ML), as well as fibrinogen (FibTEM) CT and MCF. Compared to institutional reference intervals, dogs with haemoabdomen showed prolongation of ExTEM and FibTEM CT, ExTEM CFT and 50% were hypocoagulable and 62% thrombocytopenic. No hyperfibrinolysis could be detected. Multiple linear regression models showed an association between decreased base excess, trauma and ROTEM signs for hypocoagulability. Furthermore, age was associated with a stronger fibrin clot. In conclusion, 50% of the dogs presented hypocoagulable and changes in ROTEM parameters are similar to those seen with consumption coagulopathy. Base excess and trauma were associated with hypocoagulability, while increasing age was associated with a stronger fibrin clot.

## Key words

Canine, haemoabdomen, haemostasis, hyperfibrinolysis, hypocoagulability

## Introduction

Haemoperitoneum is defined as an accumulation of blood in the abdomen. In dogs, two main reasons can be distinguished: spontaneous haemoperitoneum, caused foremost by pathological changes in the spleen or liver and traumatic haemoperitoneum due to blunt or penetrating trauma.<sup>7,28</sup>

Severe bleeding and haemorrhagic shock with or without co-occurring trauma can lead to coagulopathies that are associated with increased mortality.<sup>5</sup> Massive haemorrhage leads to both loss and consumption of coagulation factors, fibrinogen and thrombocytes.<sup>25</sup> Injury to the vascular system activates haemostasis through exposure of collagen to blood, leading to platelet adhesion and activation. Simultaneously, tissue factor is exposed and initiates the clotting cascade that subsequently leads to thrombin formation.<sup>43</sup> Excessive thrombin formation may result in thrombosis followed by a consumption coagulopathy which is similar to disseminated intravascular coagulation (DIC) and can occur with or without hyperfibrinolysis.<sup>16,18</sup> Fibrinolysis-enhanced DIC is characterized by the activation of coagulation (thrombin formation), consumption coagulopathy, and increased fibrinolysis and leads to hypocoagulability.<sup>18</sup>

In people, abdominal haemorrhage is often associated with trauma, leading to acute traumatic coagulopathy.<sup>8</sup> Hypoperfusion-associated generation of thrombomodulin-thrombin formation activates protein C, which inhibits plasminogen activator inhibitor-1 and results in hyperfibrinolysis and depletion of fibrinogen.<sup>10</sup> Regardless whether consumption or trauma-induced coagulopathy predominates, massive bleeding leads to hypocoagulability and hyperfibrinolysis.<sup>25</sup>

The incidence and underlying pathomechanisms of coagulopathy in dogs with haemoperitoneum may be similar to those identified in people but are not well described. Fletcher et al.<sup>15</sup> investigated thromboelastographic (TEG) profiles and coagulation factors of

dogs with spontaneous haemoperitoneum and described evidence of hypocoagulability, protein C deficiency and hyperfibrinolysis.

Viscoelastic blood tests such as rotational thromboelastometry (ROTEM) allow the evaluation of haemostasis from clot formation to clot dissolution in a whole blood sample. With ROTEM, the viscoelastic properties of whole blood are measured under low shear conditions, are graphically depicted in a profile and several parameters are measured and calculated by the ROTEM device. Specific coagulation activators are available that allow evaluation of both ex- and intrinsically activated haemostasis (ExTEM and InTEM assay, respectively) and evaluation of the fibrin component of the clot (FibTEM test which has cytochalasin D, a platelet inhibitor, added). Furthermore, ROTEM analysis is used to describe hyperfibrinolysis in people<sup>35,40</sup> and has been used to describe it in dogs and cats.<sup>36,37</sup>

The goal of this study was to describe haemostatic changes in dogs presenting with haemoperitoneum by means of ROTEM analysis and to evaluate associations between clinical and laboratory parameters at presentation with ROTEM parameters. We hypothesized that the dogs would show signs of hypocoagulability and hyperfibrinolysis and that these changes would correlate with the degree of hypoperfusion.

## **Material and methods**

Clinical records were searched for dogs that have been presented at the Small Animal Clinic of the Vetsuisse Faculty, University of Zurich, Switzerland, with a haemoperitoneum and for whom a ROTEM analysis at presentation was carried out. The search covered the period of five years (September 2013 – January 2019).

A haemoperitoneum was defined based on an abdominal effusion with either a packed cell volume (PCV)  $> 20\%$ <sup>28</sup> or an abdominal PCV exceeding the venous PCV in anaemic patients, or identification of a haemorrhagic source during surgery. Dogs that were treated with fluids other than an isotonic crystalloid or with drugs interfering with coagulation

(tranexamic acid, vitamin K, non-steroidal anti-inflammatory drugs, antiplatelet drugs or toxins such as rodenticides) prior to ROTEM analysis were excluded from the study.

For each dog breed, age, sex, weight and cause of the haemoperitoneum and amount of crystalloid fluid prior to ROTEM analysis were retrieved from patient records. The following clinical and laboratory parameters at admission were recorded: heart rate (HR), rectal temperature, systolic arterial blood pressure (SAP), mean arterial blood pressure (MAP), glucose, lactate and serum albumin concentration, PCV (venous and abdominal), ionized calcium (iCa), pH and base excess. The shock index was calculated from  $HR / SAP$ .<sup>24</sup>

Biochemical parameters were compared to institutional reference intervals (RI).

All ROTEM tracings were checked visually and excluded if an artefact was suspected. The ROTEM analysis was performed using citrated whole blood stored for 10-20 minutes at 37°C and followed the clinic-specific protocol, which is based on manufacturer guidelines (Tem Innovations GmbH München) and published guidelines.<sup>14</sup> The following parameters were extracted from the ROTEM database and copied into a spreadsheet for further analysis: clotting time (CT), which is defined as the time from activation until the fibrin formation starts. The clot formation time (CFT) characterizes the kinetics of clot formation and is defined as the time from start of fibrin formation until a clot of 20mm is reached. The maximum clot firmness (MCF) shows the absolute strength of the clot and is given in mm. Furthermore, maximum lysis (ML) was analysed to describe how many percent of the clot is lysed at 60 minutes. A non-measurable FIBTEM-MCF (“green line”) was defined as 0 mm. If MCF in any of the profiles did not reach 20mm, CFT was defined as 3600 seconds, as all profiles were run for 60 minutes.

Results were compared to institutional reference intervals (RI) generated from 49 dogs (Jud et al., manuscript in preparation). Hypo- or hypercoagulability was defined as at least two of the following ROTEM parameters being hypocoagulable or hypercoagulable, respectively:

ExTEM-CT, -CFT, -MCF and FibTEM-MCF. Hypofibrinogenemia was defined as FibTEM-MCF < 2 mm.

Treatment following ROTEM analysis was retrospectively extracted from patient records.

The need for surgery, transfusion requirements, the length of the ICU stay and survival (discharge from the hospital) were recorded.

Statistical analysis was carried out using IBM SPSS Statistics® software (SPSS, version 23, SPSS Inc, Chicago, IL). Data was tested for normal distribution using the Kolmogorov-Smirnov test. Normally distributed data is presented as mean  $\pm$  standard. Was the data nonnormally distributed, it is reported as median and range. To evaluate the relationship between the individual clinical parameters and ROTEM parameters (ExTEM-CT, -CFT, -MCF, -ML and FibTEM-CT and -MCF), multiple linear regression analysis was performed. Results of the multiple linear regression analysis, in combination with parameters that were significantly different from RI's, were then used to find any independent associations of clinical/biochemical parameters with the above ROTEM parameters. For those parameters that interfere with each other (HR, SAP, SI and lactate, pH, base excess) the parameter showing the strongest power in the linear regression analysis was chosen. Parameters included in the multiple linear regression model consisted of sex, age, cause of the haemoperitoneum, heart rate, temperature, glucose, venous PCV, iCa, base excess and the amount of crystalloid fluids administered prior to ROTEM analysis. A P-value < 0.05 was considered significant for all statistical analysis.

## Results

A total of 40 dogs were identified and included for analysis. Breeds included mixed breed dogs (n=6), Labrador Retriever (n=3), Bernese Mountain Dog (n=3), Boxer (n=3), Airedale Terrier (n=2), Cocker Spaniel (n=2) and Giant Schnauzer (n=2). The remaining dogs

belonged to various other breeds with on individuum per breed. Mean age of the dogs was  $8.4 \pm 3$  years (range: 1.2 – 13.4 years) and mean weight  $29.7 \pm 13.5$  kg (range: 6.5 – 69 kg).

Twelve of 40 dogs (30%) were male intact, 10/40 (25%) female, 13/40 (32.5%) male neutered and 5/40 (12.5%) female neutered.

The main cause of haemoperitoneum was spontaneous bleeding (31/40, 77.5%) due to a splenic (18/31) or hepatic mass (4/31) or other bleeding processes (9/31) such as pathological changes in the adrenal glands (3/9) or ovaries (2/9) and unknown causes of bleeding (4/9).

Nine of 40 (22.5%) dogs were identified with traumatic haemoperitoneum. Sixteen of the 40 (40%) dogs received crystalloid fluids (mean: 9 ml/kg, range: 0 – 58 ml/kg) prior to ROTEM analysis.

Clinical and laboratory parameters at the time of admission are summarized in Table 1.

ROTEM results are summarized in Table 2. Mean ExTEM-CT and -CFT and FibTEM-CT were longer than the RI. For all other analysed ROTEM parameters the mean / median was within institutional reference intervals (Table. 2). The multiple linear regression model showed that a more negative base excess was associated with a prolonged ExTEM-CT ( $t = -2.19$ ,  $p = 0.04$ ,  $\beta = -0.5$ ) and CFT ( $t = -2.48$ ,  $p = 0.02$ ,  $\beta = -0.5$ ) and FibTEM-CT ( $t = -2.42$ ,  $p = 0.03$ ,  $\beta = -0.6$ ) (Table 3). The presence of a trauma was significantly associated with a prolonged ExTEM-CFT ( $t = 2.25$ ,  $p = 0.04$ ,  $\beta = 0.40$ ). All 8 dogs that showed a reduced ExTEM-MCF were thrombocytopenic and had a reduced FibTEM-MCF. Additionally, elevated blood glucose level were associated with a shortened ExTEM-CT ( $t = -2.83$ ,  $p = 0.01$ ,  $\beta = -0.63$ ) and FibTEM-CT ( $t = -3.07$ ,  $p = 0.01$ ,  $\beta = -0.73$ ) and elevated age was associated with increased FibTEM-MCF ( $t = 2.18$ ,  $p = 0.04$ ,  $\beta = 0.60$ ).

Surgery was performed in 29/40 (72.5%) cases while the remaining 11/40 (27.5%) were treated medically. Seventeen dogs (42.5%) received a blood transfusion, including 10/40 (25%) dogs receiving erythrocyte concentrate, 2/40 (5%) an autotransfusion of abdominal whole blood, 3/40 (7.5%) fresh whole blood and 12/40 (30%) receiving fresh frozen plasma.

Mean intensive care unit stay was  $54.7 \pm 47$  hours. Thirty of 40 dogs (75%) could be discharged from the hospital, while 3 surgically and 7 medically treated dogs (25%) were euthanized.

## Discussion

The present study describing ROTEM parameters in dogs presenting with haemoperitoneum identified a prolongation of ExTEM- and FibTEM-CT and prolonged ExTEM-CFT.

Decreased base excess and trauma were associated with ROTEM signs for hypocoagulability and increased blood glucose concentration and older age were associated with signs for hypercoagulability.

The incidence of hypocoagulability in this study population was 50%. A previous study comparing dogs with spontaneous haemoperitoneum to a matched control group concluded that dogs with spontaneous haemoperitoneum were hypocoagulable based on thrombocytopenia, prolonged prothrombin time (PT) and activated partial thromboplastin time (aPTT) and a low maximal amplitude in tissue factor activated TEG.<sup>15</sup> The prolonged plasmatic coagulation times identified by Fletcher et al.<sup>15</sup> are supported by our study that identified one third of the dogs with prolonged ROTEM clotting times and mean ExTEM-CT and FibTEM-CT above the RI, as ExTEM-CT correlates to PT.<sup>11</sup> The clotting time (CT) describes the time from activation of the coagulation cascade until the amplitude of the clot reaches 2 mm and represents the time until fibrin polymerisation starts.<sup>38</sup> ExTEM- and FibTEM-CT both rely on the same coagulation factors and both tests are similarly activated. The CT depends on the strength of the activating trigger and the concentration of coagulation factors.<sup>39</sup> Thus, ExTEM- and FibTEM-CT prolongation identified in our study is thought to be a sign of decreased concentrations of coagulation factors.

Furthermore, our study identified one third of dogs with a prolonged CFT (with mean ExTEM-CFT above the RI) and a low FibTEM-MCF. The maximum clot formation in the

FibTEM represents the fibrin clot and correlates with fibrinogen concentration.<sup>11</sup> Clot formation time describes the initial rate of fibrin polymerization and depends on thrombocyte concentration, plasma fibrinogen levels and the amount of coagulation factors.<sup>42</sup> While median thrombocyte concentration was in the RI, thrombocyte concentration was decreased in two thirds of our dogs and may be causing the CFT prolongation. In addition to the prolonged clotting times, prolonged CFT, thrombocytopenia and hypofibrinogenemia (seen as low FibTEM-MCF in 14/38 dogs) support the pathomechanism of consumption coagulopathy.<sup>4,17,18,21,34</sup> In contrast to the previous study that identified decreased clot amplitude in dogs with haemoperitoneum compared to control dogs<sup>15</sup>, in the present study only 8 of 40 dogs showed decreased ExTEM-MCF and the reduced MCF was attributed to thrombocytopenia and hypofibrinogenemia.

Dilution coagulopathy, caused by extensive fluid therapy, can present similarly.<sup>29,33</sup> Dilutional coagulopathy may have influenced our results, however, our analysis showed no statistically significant association between the amount of fluid therapy prior to ROTEM analysis and the ROTEM parameters and we conclude that dogs with haemoperitoneum show signs of consumption coagulopathy.

Base excess, an indicator for systemic hypoperfusion<sup>6</sup>, was significantly associated with prolongations of ExTEM-CT, -CFT and FibTEM-CT. This indicates that hypoperfusion in patients with haemoperitoneum may be one of the causes of CT and CFT prolongation. The endothelium is very sensitive to ischemia<sup>3</sup> and reacts to it by upregulating thrombomodulin expression and increasing thrombin generation.<sup>12</sup> Thus, hypoperfusion is believed to start a cascade of events leading to various changes in haemostasis. Those changes include thrombin formation followed by consumption coagulopathy as well as activation of protein C leading to hyperfibrinolysis; these mechanisms are associated with acute traumatic coagulopathy.<sup>18,45</sup> A relation between hypoperfusion and hyperfibrinolysis has been identified in a previous study.<sup>15</sup> In contrast to that study, which used an in vitro tissue plasminogen activator activated



TEG test for detection of hyperfibrinolysis<sup>15</sup> and previous studies describing hyperfibrinolysis in dogs with haemoperitoneum.<sup>15,37,46</sup>, none of the dogs in our study showed hyperfibrinolysis. ROTEM is considered the gold standard for the identification of hyperfibrinolysis in people<sup>22,35,40</sup> and ROTEM was able to identify hyperfibrinolysis in dogs with other disease processes.<sup>26,36,37</sup> In the present study, the incidence either seems to be low or hyperfibrinolysis is mild, requiring in vitro activation with tissue plasminogen activator as used by Fletcher et al.<sup>15</sup>

Despite the changes found in ROTEM parameters supporting consumption coagulopathy rather than acute traumatic coagulopathy, we identified an association between trauma as the cause of haemoperitoneum and prolongation of ExTEM-CFT. Trauma leads to increased activation of the thrombomodulin and protein C pathways, resulting in the suppression of coagulation factors and hyperfibrinolysis.<sup>10,18</sup> Acute traumatic coagulopathy in people is defined by reductions in clot strength and hyperfibrinolysis with only limited prolongation of clotting times<sup>9</sup>. However, recent studies in dogs and cats have identified prolonged CT or plasmatic coagulation times and CFT early after trauma<sup>19,20,26,27</sup>, underlying our findings of a link between trauma and CFT prolongation. As the number of dogs presenting with traumatic haemoperitoneum was small, further studies are required to investigate the pathomechanism associated with traumatic haemoperitoneum in dogs.

Fletcher et al.<sup>15</sup> reported both hypo- and hyperfibrinogenemic dogs, which corresponds to our findings. Fibrinogen concentrations correlate with FibTEM-MCF<sup>11</sup>, which was reduced in one third of our dogs and increased in 10.5%. Older dogs of our study population presented with a higher FibTEM-MCF than younger ones. People and dogs develop alterations in the coagulation system that lead to a hypercoagulable and an antifibrinolytic state with increased age.<sup>2,13,23</sup> The development of low-grade inflammation with advancing age leads to an increased plasma fibrinogen, pro-inflammatory cytokines, and globulin levels (Ferrucci et al. 2005) and may have manifested in a higher FibTEM-MCF in our dogs.

Interestingly, we identified a significant association between blood glucose concentration and ExTEM-CT as well as FibTEM-CT shortening. Hyperglycaemia is a known phenomenon in dogs with haemorrhagic shock<sup>31</sup> and was present in 48.6% of the dogs in our study. In people, hyperglycaemia activates coagulation.<sup>30,32,41,44</sup> Despite the significant association, none of the dogs presented with a CT below the RI. If age and hyperglycaemia have a protective effect against hypocoagulopathy in dogs with haemoperitoneum remains to be determined.

There are several limitations to this study: only dogs that had ROTEM analysis performed were included, therefore the study needs to be interpreted with respect to its population. Dogs receiving isotonic crystalloid fluids were not excluded and while multiple linear regression analysis did not identify fluid therapy as a significant factor contributing to the prolonged CT and CFT, a dilutional effect cannot completely be excluded. Additionally, the data was compared to institutional RI derived for the machine and were not compared to a matched control group. Furthermore, being a clinical study, the amount and timeframe of blood loss is not known and as haemostasis is a dynamic process, the results may vary under different conditions. To obtain a more detailed insight into pathomechanisms associated with canine haemoperitoneum, further studies with prospective designs are needed.

In conclusion, the changes in ROTEM parameters in this clinical study of dogs with both spontaneous and traumatic haemoperitoneum are similar to those seen with consumption coagulopathy and include CT and CFT prolongation, thrombocytopenia and low FibTEM-MCF leading to 50% of the dogs presenting hypocoagulable. Base excess and trauma were associated with hypocoagulability, while increasing age was associated with a stronger fibrinogen clot.

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Table 1: Clinical and laboratory parameters at presentation of 40 dogs with haemoperitoneum

Parameter	N	Abnormal dogs n (%)	Mean $\pm$ SD Median	Minimum	Maximum	Reference Interval RI <sup>1</sup>
Heart Rate (beats per minute)	39	↑ 27 (69.2%)	139	68	200	60 - 120
Systolic arterial pressure (mmHg)	33	↑ 8 (24.2%) ↓ 10 (30.3%)	108	60	150	100 – 150
Mean arterial pressure (mmHg)	35	↓ 8 (22.9%)	68	40	118	60 - 120
Shock Index	33	↑ 29 (87.8%)	1.23 $\pm$ 2.55	0.53	3.08	< 1.0
Temperature (°C)	38	↑ 4 (10.5%) ↓ 13 (34.2%)	37.7 $\pm$ 0.9	35.8	39.6	37.5 - 39.0
Lactate (mmol/L)	34	↑ 20 (58.8%) ↓ 1 (2.9%)	2.87	0.36	16.80	0.42-2.13
pH	33	↓ 19 (57.6%)	7.304 $\pm$ 0.116	6.900	7.440	7.35 - 7.45
Base excess (mmol/L)	33	↓ 18 (54.5%)	-6.30 $\pm$ 4.75	-17.6	0.3	-5 – 2

PCV peripheral (%)	39	↓ 36 (92.3%)	29 ± 8	16	57	37 -61
PCV abdominal (%)	29	–	33 ± 10	12	60	–
Thrombocyte count (10 <sup>3</sup> /μl)	24	↓ 15 (62.5%)	131 ± 95	26	411	> 130
Glucose (mmol/L)	37	↑ 18 (48.6%) ↓ 1 (2.5%)	7.57 ± 2.95	3.2	17.2	3.9-6.6
iCa (mmol/L)	33	↓ 10 (25.6%)	1.21 ± 0.07	1.0	1.35	1.21 – 1.41

iCa, ionized calcium; PCV, packed cell volume; SD, standard deviation; ↑, number of dogs above the RI; ↓, number of dogs below the RI  
 results that deviate from the reference interval are marked in bold

Table 2: ROTEM parameters of 40 dogs presenting with haemoperitoneum

Coagulation parameter		N	Abnormal dogs n (%)	Mean $\pm$ SD Median	Minimum	Maximum	RI
ExTEM	CT (sec)	40	<b><math>\uparrow</math> 14 (35%)</b>	<b>93* <math>\pm</math> 48</b>	23	174	23 – 87
	CFT (sec)	40	<b><math>\uparrow</math> 14 (35%)</b> <b><math>\downarrow</math> 1 (2.5%)</b>	<b>382* <math>\pm</math> 367</b>	18	2024	85 – 357
	MCF (mm)	40	<b><math>\uparrow</math> 2 (5%)</b> <b><math>\downarrow</math> 8 (20%)</b>	42	20	80	32 – 65
	ML (%)	40		4 $\pm$ 4	0	11	0 – 12
FibTEM	CT (sec)	38	<b><math>\uparrow</math> 15 (39.5%)</b>	<b>941* <math>\pm</math> 1330</b>	21	3600	21 – 112
	MCF (mm)	38	<b><math>\uparrow</math> 4 (10.5%)</b> <b><math>\downarrow</math> 4 (10.5%)</b>	5 $\pm$ 8	0	45	2 – 9

CFT, clot formation time; CT, clotting time; MCF, maximal clot formation; ML, maximal lysis; RI, reference interval; SD, standard deviation;  **$\uparrow$** ,

number of dogs above the RI;  **$\downarrow$** , number of dogs below the RI

results that deviate from the reference interval are marked in bold

10 Table 3: multiple linear regression analysis of clinical and biochemical parameters compared to ROTEM parameters in 40 dogs with haemoperitoneum

ROTEM parameter		Sex	Age	Cause	Heart rate	Temperature	Glucose	PCV peripheral	iCa	BE	Crystalloid fluids
ExTEM	CT	t = 0.28 p = 0.78	t = 0.94 p = 0.36	t = 0.5 p = 0.62	t = 1.51 p = 0.15	t = 0.17 p = 0.87	t = -2.83 p = 0.01 $\beta$ = -0.63	t = 0.38 p = 0.71	t = -0.32 p = 0.75	t = -2.19 p = 0.04 $\beta$ = -0.50	t = -0.96 p = 0.35
	CFT	t = 0.29 p = 0.77	t = 0.43 p = 0.67	t = 2.25 p = 0.04 $\beta$ = 0.40	t = 1.33 p = 0.2	t = 1.84 p = 0.08	t = -1.76 p = 0.09	t = 1.04 p = 0.31	t = -0.35 p = 0.73	t = -2.48 p = 0.02 $\beta$ = -0.50	t = -1.42 p = 0.17
	MCF	t = -0.1 p = 0.92	t = 0.25 p = 0.81	t = -1.11 p = 0.28	t = -0.86 p = 0.4	t = -0.86 p = 0.56	t = 0.6 p = 0.56	t = 0.04 p = 0.97	t = 1.21 p = 0.24	t = 0.61 p = 0.55	t = 0.94 p = 0.36
	ML	t = -1.44 p = 0.16	t = 2.00 p = 0.06	t = 2.00 p = 0.23	t = -2.07 p = 0.05	t = -1.12 p = 0.56	t = 0.04 p = 0.97	t = 0.34 p = 0.74	t = -1.01 p = 0.32	t = -1.2 p = 0.24	t = 1.56 p = 0.13
FibTEM	CT	t = -0.91 p = 0.38	t = -0.01 p = 0.99	t = -0.01 p = 0.99	t = 1.68 p = 0.11	t = -1.7 p = 0.11	t = -3.07 p = 0.01 $\beta$ = -0.73	t = -1.37 p = 0.19	t = 0.84 p = 0.41	t = -2.42 p = 0.03 $\beta$ = -0.60	t = -0.4 p = 0.69

	MCF	$t = 0.77$	$t = 2.18$	$t = -0.71$	$t = -0.66$	$t = -0.64$	$t = -0.46$	$t = 1.5$	$t = 0.39$	$t = -0.4$	$t = 1.3$
	$R^2 = 0.278$	$p = 0.45$	$p = 0.04$ $\beta = 0.60$	$p = 0.49$	$p = 0.52$	$p = 0.53$	$p = 0.65$	$p = 0.15$	$p = 0.7$	$p = 0.7$	$p = 0.21$

iCa, ionized calcium; BE, base excess; CFT, clot formation time; CT, clotting time; MCF, maximal clot formation; ML, maximal lysis; PCV, packed cell volume;  $R^2$  = adjusted R-Square;  $t$  = T-Test;  $\beta$  =standardized beta coefficient;  
 numbers in bold show statistically significant association

## Table legends

Table 1: Clinical and laboratory parameters at presentation of 40 dogs with haemoperitoneum iCa, ionized calcium; PCV, packed cell volume; SD, standard deviation; ↑, number of dogs above the RI; ↓, number of dogs below the RI

results that deviate from the reference interval are marked in bold

Table 2: ROTEM parameters of 40 dogs presenting with haemoperitoneum CFT, clot formation time; CT, clotting time; MCF, maximal clot formation; ML, maximal lysis; RI, reference interval; SD, standard deviation; ↑, number of dogs above the RI; ↓, number of dogs below the RI

results that deviate from the reference interval are marked in bold

Table 3: multiple linear regression analysis of clinical and biochemical parameters compared to ROTEM parameters in 40 dogs with haemoperitoneum iCa, ionized calcium; BE, base excess; CFT, clot formation time; CT, clotting time; MCF, maximal clot formation; ML, maximal lysis; PCV, packed cell volume;  $R^2$  = adjusted R-Square; t = T-Test;  $\beta$  =standardized beta coefficient; numbers in bold show statistically significant association

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